

1 1. A purified and isolated nucleic acid molecule
2 comprising a nucleic acid sequence selected from the
3 group consisting of (1) the porcine nucleic acid sequence
4 depicted in Figure 4 (SEQ ID NO: 7), (2) a sequence
5 corresponding to the sequence of (1) within the scope of
6 the degeneracy of the genetic code, (3) a sequence that
7 encodes a porcine polypeptide having α -1,3
8 galactosyltransferase activity and that hybridizes under
9 standard high stringency conditions with a sequence
10 complementary to the sequence of (1) or (2), and (4) a
11 sequence complementary to the sequence of (1), (2) or
12 (3).

1 2. A host cell that is transformed with the
2 nucleic acid molecule of claim 1.

1 3. A porcine α -1,3 galactosyltransferase encoded
2 by the nucleic acid molecule of claim 2.

1 4. A DNA construct useful for inactivating the
2 porcine α -1,3 galactosyltransferase gene by insertion of
3 a desired DNA sequence into an insertion site of said
4 gene, comprising said desired DNA sequence flanked by
5 first and second homology sequences, said first and
6 second homology sequences being, respectively,
7 sufficiently homologous to first and second genomic
8 sequences flanking said insertion site to allow for
9 homologous recombination of said DNA construct with said
10 porcine α -1,3 galactosyltransferase gene when said DNA
11 construct is introduced into a porcine cell having said
12 α -1,3 galactosyltransferase gene.

1 5. The DNA construct of claim 4, wherein said
2 insertion site is within exon 4, exon 7, exon 8 or exon 9
3 of the porcine α -1,3 galactosyltransferase gene.

1 6. The DNA construct of claim 4, wherein said
2 desired DNA sequence is selected from the group
3 consisting of the neo^R gene, the hyg^R gene and the
4 thymidine kinase gene.

1 7. The DNA construct of claim 6, wherein said
2 desired DNA sequence is bordered at the 5' and 3' ends by
3 FRT DNA elements, and wherein stop codons for each of the
4 three reading frames have been inserted 3' to the desired
5 DNA sequence.

1 8. A DNA construct useful for inactivating the
2 murine α -1,3 galactosyltransferase gene by insertion of a
3 desired DNA sequence into an insertion site of said gene,
4 comprising said desired DNA sequence flanked by first and
5 second homology sequences, said first and second homology
6 sequences being, respectively, sufficiently homologous to
7 first and second genomic sequences flanking said
8 insertion site to allow for homologous recombination of
9 said DNA construct with said murine α -1,3
10 galactosyltransferase gene when said DNA construct is
11 introduced into a murine cell having said α -1,3
12 galactosyltransferase gene.

1 9. The DNA construct of claim 8, wherein said
2 insertion site is within exon 4, exon 7, exon 8 or exon 9
3 of the murine α -1,3 galactosyltransferase gene.

1 10. The DNA construct of claim 8, wherein said
2 desired DNA sequence is selected from the group
3 consisting of the neo^R gene, the hyg^R gene and the
4 thymidine kinase gene.

1 11. The DNA construct of claim 10, wherein said
2 desired DNA sequence is bordered at the 5' and 3' ends by
3 FRT DNA elements, and wherein stop codons for each of the
4 three reading frames have been inserted 3' to the desired
5 DNA sequence.

1 12. A method for generating a mammalian totipotent
2 cell having at least one inactivated α -1,3
3 galactosyltransferase allele, said totipotent cell
4 derived from a mammalian species having a functional α -
5 1,3 galactosyltransferase gene, comprising:
6 (a) providing a plurality of cells characterized as
7 totipotent cells of said mammalian species;
8 (b) introducing into said totipotent cells a nucleic
9 acid construct effective for inactivating said α -1,3
10 galactosyltransferase gene by insertion of a desired DNA
11 sequence into an insertion site of said gene through
12 homologous recombination; and
13 (c) identifying a totipotent cell having at least
14 one inactivated α -1,3 galactosyltransferase allele.

1 13. The method of claim 12 in which said totipotent
2 cell is a murine ES cell.

1 14. The method of claim 12 in which said totipotent
2 cell is a murine egg.

1 15. The method of claim 12 in which said totipotent
2 cell is a porcine ES cell.

1 16. The method of claim 12 in which said totipotent
2 cell is a porcine PGC.

1 17. The method of claim 12 in which said totipotent
2 cell is a porcine egg.

1 18. A method for generating a mammal lacking a
2 functional α -1,3 galactosyltransferase gene, said mammal
3 belonging to a species having a functional α -1,3
4 galactosyltransferase gene, comprising:

5 (a) providing a mammalian totipotent cell having at
6 least one inactivated α -1,3 galactosyltransferase allele,
7 said totipotent cell derived from a mammalian species
8 having a functional α -1,3 galactosyltransferase gene;

9 (b) manipulating said totipotent cell such that
10 mitotic descendants of said cell constitute all or part
11 of a developing embryo;

12 (c) recovering a neonate derived from said embryo;
13 and

14 (d) raising and breeding said neonate to obtain a
15 mammal homozygous for said inactivated α -1,3
16 galactosyltransferase allele.

1 19. The method of claim 18, wherein said totipotent
2 cell is a murine ES cell and said manipulating comprises
3 injecting said ES cell into the blastocyst cavity of a
4 murine blastocyst and implanting said injected blastocyst
5 into a murine recipient female.

1 20. The method of claim 18, wherein said totipotent
2 cell is a murine egg, and said manipulating comprises
3 implanting said egg into a murine recipient female.

1 21. The method of claim 18, wherein said totipotent
2 cell is a porcine ES cell and said manipulating comprises
3 injecting said ES cell into the blastocyst cavity of a
4 porcine blastocyst and implanting said injected
5 blastocyst into a porcine recipient female.

1 22. The method of claim 18, wherein said totipotent
2 cell is a porcine ES cell and said manipulating comprises
3 injecting said ES cell into a porcine morula.

1 23. The method of claim 18, wherein said totipotent
2 cell is a porcine ES cell and said manipulating comprises
3 co-culture of said ES cell with a zona pellucida-
4 disrupted porcine morula.

1 24. The method of claim 18, wherein said totipotent
2 cell is a porcine ES cell and said manipulating comprises
3 fusing said ES cell with an enucleated porcine zygote.

1 25. The method of claim 18, wherein said totipotent
2 cell is a porcine egg, and said manipulating comprises
3 implanting said egg into a porcine recipient female.

1 26. A mammal lacking a functional α -1,3
2 galactosyltransferase gene, said mammal belonging to a
3 species having a functional α -1,3 galactosyltransferase
4 gene, said mammal produced by the method of claim 18.

1 27. The mammal of claim 26, wherein said mammal is
2 a mouse.

1 28. The mammal of claim 26, wherein said mammal is
2 a pig.

1 29. A non-naturally occurring mammal lacking a
2 functional α -1,3 galactosyltransferase gene, said mammal
3 belonging to a species having a functional α -1,3
4 galactosyltransferase gene.

1 30. The mammal of claim 29, wherein said mammal is
2 a mouse.

1 31. The mammal of claim 29, wherein said mammal is
2 a pig.

1 32. A purified and isolated nucleic acid molecule
2 comprising a nucleic acid sequence selected from the
3 group consisting of (1) the nucleic acid sequence
4 depicted in Figure 26 (SEQ ID NO: 25), (2) a sequence
5 corresponding to the sequence of (1) within the scope of
6 the degeneracy of the genetic code, (3) a sequence that
7 encodes murine T-LIF and that hybridizes under standard
8 high stringency conditions with a sequence complementary
9 to the sequence of (1) or (2), and (4) a sequence
10 complementary to the sequence of (1), (2) or (3).

1 33. A host cell that is transformed with the
2 nucleic acid molecule of claim 32.

1 34. A murine T-LIF polypeptide encoded by the
2 nucleic acid molecule of claim 32.

1 35. A purified and isolated nucleic acid molecule
2 comprising a nucleic acid sequence selected from the
3 group consisting of (1) the nucleic acid sequence
4 depicted in Figure 27 (SEQ ID NO: 31), (2) a sequence
5 corresponding to the sequence of (1) within the scope of
6 the degeneracy of the genetic code, (3) a sequence that
7 encodes human T-LIF and that hybridizes under standard
8 high stringency conditions with a sequence complementary
9 to the sequence of (1) or (2), and (4) a sequence
10 complementary to the sequence of (1), (2) or (3).

1 36. A host cell that is transformed with the
2 nucleic acid molecule of claim 35.

1 37. A human T-LIF polypeptide encoded by the
2 nucleic acid molecule of claim 35.

1 38. A method for eliminating or reducing hyperacute
2 rejection of non-primate mammalian cells, tissues and
3 organs by human serum, comprising adding, to said human
4 serum, a physiologically acceptable amount of galactose
5 or a saccharide in which the terminal carbohydrate is an
6 α galactose linked at position 1, prior to exposure of
7 said human serum to said non-primate cells, wherein said
8 amount of galactose or saccharide is sufficient to reduce
9 or eliminate said hyperacute rejection.

1 39. The method of claim 38, wherein said saccharide
2 is selected from the group consisting of melibiose,
3 galactose α 1-3 galactose and stachyose.

1 40. A method for eliminating or reducing hyperacute
2 rejection of non-primate mammalian cells, tissues and
3 organs by human serum, comprising substantially depleting
4 said serum of immunoglobulin.

1 41. A method for eliminating or reducing hyperacute
2 rejection of non-primate mammalian cells, tissues and
3 organs by human serum, comprising substantially depleting
4 said serum of IgM antibodies.

1 42. A method for eliminating or reducing hyperacute
2 rejection of non-primate mammalian cells by human serum,
3 comprising substantially depleting said serum of anti-GAL
4 IgM and IgG antibodies.

1 43. A method for eliminating or reducing hyperacute
2 rejection of non-primate mammalian cells by human serum,
3 comprising substantially depleting said serum of anti-GAL
4 IgM antibodies.

1 44. Affinity-treated human serum substantially free
2 of anti-GAL antibodies.

1 45. Affinity-treated human serum substantially free
2 of anti-GAL IgM antibodies.